Pineapple News

Newsletter of the Workgroup Pineapple, International Society for Horticultural Science
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https://docs.google.com/spreadsheets/d/1ePrvbOxZK_fAetf5dDG9KSJ-iI3TGbWEuVPqNWSIC1k/pubhtml

Workgroup Pineapple News

IX International Pineapple Symposium (IPS) proceedings

The proceedings of the symposium, Acta Horticulturae Number 1239, was published in April 2019. The book contains 25 articles in 212 pages. A list of the papers and copies of the book are available at https://www.actahort.org/books/1239/.

X International Pineapple Symposium (IPS)

The X International Pineapple Symposium is scheduled to be held in Punta Cana, Dominican Republic, on 22-26 April 2020, Convener: Mr. Joelin Santos. Please see additional details below under N**ews from Dominicum Republic**.

Links to information on pineapple.

Py, C., Lacoeuilhe, J.J., and Teisson, C. 1984. **L'Ananas Sa Culture, Ses Proudits**. Paris, Editions G.-P. Maisonneuv,

Py, C., Lacoeuilhe, J.J. & Teisson, C. 1987) **The pineapple. Cultivation and uses**, Paris, Editions G.-P. Maisonneuve are available at https://evols.library.manoa.hawaii.edu/handle/10524/55419.

Sanewski, G.M., Bartholomew, D.P. and Paull, R.E. & 20 other international experts. 2018. **The Pineapple: Botany, Production and Uses**, 2nd edition. See Pineapple News No. 25 or https://www.cabi.org/bookshop/book/9781786393302 for more details.

The Mexican pineapple: Advances and challenges in innovation management. For details, see News from Mexico on p. 31.

ISHS Workgroup Pineapple and Pineapple News archives

See whats new at https://www.ishs.org/pineapple

All back copies of Pineapple News can be found at the links below**.**

https://www.ishs.org/pineapple/pineapple-newsletters

https://scholarspace.manoa.hawaii.edu/handle/10125/41067

A searchable table of contents of all issues of Pineapple News can be viewed at:

https://docs.google.com/spreadsheets/d/1ePrvbOxZK_fAetf5dDG9KSJ-iI3TGbWEuVPqNWSIC1k/pubhtml

Pineapple cultivar listing by country. Additions and corrections welcomed.

https://docs.google.com/spreadsheets/d/1NDr9v3FSZLP8W3m9rYhScErrxed8vEc7f8meDyLXEmg/edit?usp=sharing

The google spreadsheet provides a searchable cultivar list that is alphabetized by country and by cultivars within each country where the various cultivars are being grown. The list also includes any colloquial name used in the country, other relevant comments and the first published mention found of the cultivar in each country.

The objective of producing a listing of the world's pineapple cultivars is to increase readership of important papers and to link those papers to the body of research pertaining to that cultivar.

In addition to documenting the presence of cultivars within a country, the list can be used to identify the pineapple cultivar in papers that often only mention the colloquial (local) name of the cultivar. Some colloquial names, "Sugarloaf" and "Sugar loaf" are examples, are used to name more than one cultivar. A true cultivar has only one name.

Local names can be used in publications, but they should always be listed after the true cultivar name and using the true cultivar name will assure that research papers will reach the widest possible audience.

News from Australia

In Memorium: The Pineapple Journey of Col Scott

Garth M. Sanewski, Maroochy Research Facility, Department of Agriculture and Fisheries, Nambour. QLD. Australia. E-mail: Garth.Sanewski@daf.qld.gov.au

Many of the international pineapple community will have heard of Col Scott. Sadly, Col passed away June 23, 2018 after a lifetime of working for the Australian and briefly, the South African pineapple industries. The loss of Col and his long experience marks a generational changing of the guard. Col was appreciated for his knowledge and willingness to assist anyone even remotely connected with the industry. His guidance was keenly sought on all manner of issues and is now sadly missed. This is a brief account of Col's contribution.

Mar 1965 — Oct 1967: Col worked for Queensland Department of Primary Industries, Horticulture Branch, Ormiston (near Brisbane) as a Field Assistant, Banana Inspector (for Bunchy Top virus) and Extension Officer.

Oct 1967 — Oct 1969: Col was in National Service in the Australian Army and served as a 2nd Lt in the RAASC as a Transport Platoon Commander at Victoria Barracks, Melbourne

Mar 1970 — Sep 2002: Col worked with the processor Golden Circle Ltd, firstly as a horticulturist and then, as Horticultural Manager. This is a period where Col met and influenced many Queensland pineapple growers providing contact, support and research services. Col established new cultivars, maintained links with and made representations to government and industry. Col authored many technical articles and his expertise on pineapples was sought for articles and lectures. Col managed the establishment of 73-50 and MD-2, a Quality Assurance Program for the pineapple industry and research projects and made recommendations to the Board of Directors. Time was spent collecting industry statistics, collecting and analyzing scientific data and at all times being accessible for advice on horticultural problems and the application of the latest scientific advice.

Nov 2002 — Dec 2007: Col established himself in East London South Africa with Summerpride Foods Ltd. Col initiated a plant breeding program which produced 10,000 seedlings and sourced, bred and set up an ornamental pineapple business. Importantly, Col advised growers with horticultural advice and worked closely with growers establishing farm visits, maintaining worldwide links and provided advice on identifying new business opportunities.

Jan 2008 — May 2011: Col consulted on a private and confidential basis to individuals and groups on all facets of the production of pineapples for processing and/or fresh market.

June 2011 - April 2018: Col worked as an Agronomist with Tropical Pines Pty Ltd and was a familiar face on all farms collecting data, providing advice and encouragement, establishing trials, liaising with industry bodies and helping implement plant selection and breeding programs.

Genetic Resistance to *Phytophthora cinnamomi* in pineapple

G. Sanewski and H-L. Ko. Maroochy Research Facility, Department of Agriculture and Fisheries, Nambour. QLD. Australia.

Summary. Genetic resistance to *Phytophthora cinnamomi* in a bi-parental pineapple seedling population derived from 'MD-2' and 'PRI-59-656' is associated with a genomic region on chromosome 5 that is rich in defence genes. The most highly likely causative genes include a Jasmonate-zim domain 10 and a Spermidine/ putrescine-binding protein although polymorphisms in several LRR's, a Xyloglucan endotransglucosylase/ hydrolase and transcription factor bHLH are also associated. Linkage data for the most highly associated SNP marker implicates a range of other genes including a glycosyl hydrolase, terpenoid cyclase, CBS domain protein, mitogen-activated kinase, GTP cyclohydrolase and glutathione S-transferase could be part of the resistance response. The putative set of implicated and associated genes suggests a comprehensive response to *P. cinnamomi* by pineapple including ROS regulation, calcium signalling, cell wall modification and pathogen attack.

Introduction

Root and heart rots caused by *Phytophthora* species are a major disease of pineapple worldwide. *Phytophthora cinnamomi* (*Pc*), a hemi-biotroph (Van den Berg et al. 2018) causes root rots that in severe cases can develop into heart rot.

The centre of origin for pineapple is considered to include northern Brazil and Paraguay (Coppens de Eckenbrugge et al. 2018). *Phytophthora* originated in Taiwan (Jung et al. 2017) but speciation has included other regions. *P. cinnamomi*, while originating in Taiwan, might also have an ancient distribution throughout parts of Asia, Papua New Guinea and northern Australia (Arentz, 2017). Additional speciation of *Phytophthora*, including the potato pathogen, *P. infestans*, occurred in Central America (Shakya, 2018). It is possible therefore, that pineapple evolved with *P. infestans* and some other *Phytophthora* species, but not *P. cinnamomi*.

Resistance in pineapple exists across the spectrum of genotypes from wild to modern domestic. Immunity has not however been shown. Typically, most genotypes can be infected given the appropriate conditions but highly resistant genotypes can recognise infection, respond quickly to limit damage and subsequently, depending on genotype, produce new roots. Varietal susceptibility to infection and the ability to quickly regenerate new roots varies considerably. The popular fresh market cultivar, MD-2 is highly susceptible, possibly one of the most susceptible genotypes along with 'Manzana'. A high level of resistance exists in the Pineapple Research Institute (PRI) *Ananas comosus* var. *comosus* cultivar 'PRI 59-656' and *A. comosus* var. *bracteatus* (Sanewski et al. 2016).

In a previously published genome-wide association study (GWAS) by Sanewski et al, (2017) a population of seedlings segregating for resistance/ susceptibility was screened for resistance and sequenced for DArTseq markers. The parents included the resistant varieties 'PRI 59-656' and *A. comosus* var. *bracteatus* accession 'FRF19' and the susceptible *A. comosus* var. *microstachys* accession 'FRF223' and three domestic *A. comosus* var. *comosus* cultivars, 'MD-2', '11-149' and '22-590'. The majority of the population derived from the parents 'MD-2' and 'PRI 59-656'. Using a GWAS approach, resistance was associated with a single, broad locus between the positions 4.6-5.5 Mb on chromosome five. Thirteen genes were found associated with resistance although it is considered the association of some of these is at least partially by linkage.

Further analysis was required to understand which are the more important genes of those identified and the processes underlying their function. Here we further analyse the marker associations using additional algorithms some of which use a multi-locus approach to minimise confounding effects of kinship and linkage. This provides a shorter list of candidate genes that, in combination with genome positional data and flanking genes, provides more information on likely causative genes.

Three GWAS models were applied to the dataset using the GAPIT R software package 3 (Zhang et al. 2010; Tang et al. 2016). These include the enriched compressed mixed linear model (ECMLM), the multi-loci linear mixed model (MLMM) (Segura et al. 2012) and FarmCPU (fixed and random model circulating probability unification) (Liu et al. 2016). The first is an enhanced mixed linear single loci model and the others are multi-loci models. The ECMLM, while offering high statistical power usually cannot separate linked genes even if they are spread across the genome. Typically, this model will identify a greater number of markers associated with genes, many of which will be linked but possibly still functional. The multi-loci models initially analyse the genome in discreet regions and only include the mostly highly associated in further steps. Typically, multi-loci models identify only the more significant markers from each potential QTL. The multi-loci models, because they bin markers according to physical position, are not appropriate for unpositioned markers or those on short scaffolds. All models minimise false positives. Three covariates were used to account for population structure. The QQ plots were optimal and only one QTL, on LG05, was found significant' although some markers positioned on two scaffolds were also significant. It is expected these scaffolds should be positioned on LG05. The results for the ECMLM analysis of SNP markers are shown in table 1.

Here we take a closer look at the two most significant SNP markers.

Results

Main candidate genes

The highest associated SNP marker across the three analyses was #100029071 positioned on chromosome 5 at 4,708,927 bp. This was the only marker found significantly associated using the FarmCPU and MLMM algorithms. This SNP marker, unmatched in BLAST and hence not shown in table 1, is positioned between a Spermidine/putrescine-binding periplasmic protein and a reverse transcriptase Retrotransposon protein but only 26 kb from a jasmonate-zim-domain protein 10.

Of the genes positioned near the marker, the Jasmonate-zim-domain protein 10 and spermidine/putrescine-binding protein are considered the most important. The jasmonate-zim-domain protein 10, a transcription factor also known as TIFY9, is the main regulator of the jasmonic acid (JA) pathway (Sen et al. 2016). The jasmonic acid pathway is associated with plant response to infection by *Phytophthora* species (Allardyce et al. 2013). In this study, a silicoDArT marker (present/ absent) matching this gene was also found highly associated (data not shown). This indicates there is a functional change in the Jasmonate-zim domain protein associated with resistance/ susceptibility but it was not demonstrated to be a single nucleotide change. This is likely the most important of the gene found associated with a differential response to *Pc*.

Spermidine and putrescine are polyamines that are known to function in various aspects of stress and plant defence response, including stimulation of systemic-acquired resistance (Seifi et al. 2017) and enhancement of anti-oxidant capacity in response to stress (Yadu et al. 2018). As an example, application of spermine was shown to induce systemic resistance to a necrotrophic pathogen, *Botrytis cineara*, in tomato, bean and *Arabidopsis*. While this was shown to stimulate the hypersensitive response and reactive oxygen species, this was of a limited extent in the early stages of infection thus not favouring the necrotroph to the extent as would normally be expected (Seifi et al. 2017). Exogenously applied spermine was also shown to increase resistance to *Phytophthora capsici* in capsicum (Koc et al. 2017).

The proximity of the jasmonate and spermidine genes (27 kb) and complimentary roles suggest they might function together. The low SNP density means however, only one marker is positioned in this specific region and hence might be marking both genes. For this same reason, there is no linkage data for this gene pair. It is highly likely however, that they are in linkage disequilibrium (LD).

The second highest associated marker, #100046224, was matched in BLAST to an Uncharacterised protein. This marker is some distance, 458 kb, from the first marker but is very highly linked (R^2=0.91, D′=1.0). There are no other genes of interest, apart from a Farnesyltransferase/geranylgeranyltransferase protein, positioned immediately near this second marker and #100046224 is likely associated mostly by linkage to the main candidate marker, #100029071. Notwithstanding this, the Farnesyltransferase/geranylgeranyltransferase protein is known to be involved in plant defence response (Goritschnig et al. 2008) and, as such, is also of interest.

Linked genes of interest

As mentioned already, marker density was low in the genomic region of interest. Other functional important genes could therefore exist within this region but not be represented by a marker. Linkages and flanking positions therefore become more important. Markers linked to marker #100029071 with an R^2>0.5 are shown in table 2. While few of these linked genes were matched in BLAST (MegaBLAST), putative genes have been identified for each linked marker based on positional data in the 'Smooth Cayenne' genome v3 (Ming et al, 2015). There are seven putative linked genes that will be briefly discussed here.

- <u>Terpenoid cyclase (terpenoid synthase)/ prenyltransferase</u> superfamily have been shown to be involved in the production of phytoalexins. In particular, prenyltransferases were involved in the synthesis of phytoalexin glyceollins in soybean in response to infection by *Phytophthora sojae* (Sukumaran et al. 2018). Terpene synthases were also highly up regulated in maize in response to infection by *P. cinnamomi* and were considered the key component of resistance to *P. cinnamomi* in that species (Allardyce et al. 2013). Interestingly, the study of Allardyce et al, (2013) also associated genes in the jasmonic acid pathway as well as terpenoid synthase with response to *P. cinnamomi* infection in maize.
- <u>Glycosyl hydrolases</u> degrade long chain carbohydrates and as such are involved in cell wall metabolism including lignification, but also in defence. β glucosidases and chitinases are classes of gycosyl hydrolases (Minic, 2008; Cao et al. 2019). Chitinases are important in defence against fungal pathogens as it acts directly against the pathogen.
- In rice, a <u>CBS (cystathionine-β synthase) domain-containing protein</u> is up-regulated by inoculation with a hemi-biotrophic pathogen, Rice Blast, and also by exogenously applied salicylic acid (SA) or methyl jasmonate. It is considered a positive regulator of resistance (Mou et al. 2015). Also, AMP-activated protein kinases contain a CBS domain. These proteins are involved in oxidation within the cell.
- <u>Mitogen-activated kinases</u> (MAPKs) play a role in signal transduction through other kinases, proteins and transduction factors in response to various cues. MAPKs are involved in JA and SA signalling both as positive and negative regulators (Jagodzik et al. 2018).
- <u>GTP cyclohydrolase</u> (RibA) is a key gene in the biosynthesis of riboflavin and ascorbic acid in maize (Shan et al. 2019). Ascorbic acid is one of several ROS scavenging mechanisms that

help control the hypersensitive response (Huang et al. 2019). DHBP synthase (RibB) functions with RibA at least in the biosynthesis of riboflavin.

- Glutathione S-transferase (GST) is involved in the ascorbic acid cycle and as such performs oxygen-scavenging activities to regulate reactive oxygen species (ROS) (Acosta-Muniz et al. 2012).
- Inositol-1, 4, 5-triphosphate 5-phosphatase is involved in the phosphoinositide calcium signalling pathway. This pathway is a key part of basal and systemic-acquired resistance response to plant pathogens (Hung et al. 2014).

Co-Expressed genes

Publically available co-expression data for 'Smooth Cayenne' and 'MD-2' (http://pineapple.angiosperms.org/pineapple/html/index.html.) for various tissues, even though not from plants challenged with a pathogen, can give an insight into the possible role of some of the genes putatively involved in resistance. This data was not generated in the current study.

No co-expression data exists for the two principal causative genes, possibly due to a lack of regulation in the absence of the pathogen, but does for one of the linked genes. Glycosyl hydrolase has 81 co-expressed genes with a Pearson coefficient (+) >0.7. Data for genes where expression in the roots (FPKM) is high are shown in table 3 as an example of co-expressed genes. Collectively these are involved in regulation of ROS and the hypersensitive response, the JA pathway and pathogen attack.

Conclusion

The two principal causative candidates for *Pc* resistance in pineapple in the genetic background studied are the jasmonate zim-domain protein 10 and the spermidine/putrescine-binding periplasmic protein. These likely provide the main resistance effect in the PRI cultivar, PRI 59-656, but other genes associated, linked and co-expressed also contribute to a lesser but important extent. Potential loss-of-function variants of these genes is likely why 'MD-2' is so susceptible. Having all the genes in the correct allelic state is likely to provide the most robust resistance. The collective data on associated, linked and co-expressed genes suggests a role for the hypersensitive response and JA pathway in resistance to *P. cinnamomi*.

Only the two most highly associated SNPs have been discussed here but additional functional genes are suggested by other associated SNPs and silicoDArT (not shown) markers and their potential gene networks. Other genes of interest include Homogentisate 1,2-dioxygenase, Fibrillin, Prostaglandin synthase, Acyl-acyl-carrier protein desaturase, NADH-quinone oxidoreductase, GDSL lipase, Farnelsyltransferase, Xyloglucan endo-transglycosylase, 3beta-hydroxysteroid-dehydrogenase/decarboxylase and NAC domain protein as well as several LRRs and RKs. There is however no assertion there are polymorphisms in all of these genes.

The markers and matched genes found associated by GWAS might not explain the full repertoire of response by the pineapple plant to *Phytophthora* infection as hinted in the co-expression data. A more detailed study of differential gene expression in response to infection in resistant and susceptible genotypes is needed to better understand the relative role of each gene. An engineered loss-of-function study in a model monocot, say rice, focusing on the two main candidate genes might also be helpful.

Acknowledgements. The project was funded by Horticulture Innovation Australia Pty Ltd (Hort Innovation) using voluntary contributions from the Department of Agriculture and Fisheries and matched funds from the Australian Government.

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Table 1. SNP markers found associated with resistance/ susceptibility to *P. cinnamomi* in pineapple using the ECMLM algorithm in GAPIT 3 and matched to genes in BLAST. (MAF=minor allele frequency).

Marker	Position Gene ID	-Log10 (P) Corrected	Length (bp)	MAF	Gene	% Cover/ identity	BLAST E value
100046224	LG05: 5,166,727	7.3	69	0.47	*Ananas comosus* uncharacterised (LOC109710562) Transcript variants X1-6	95/ 99	6e -25
4720783*	LG05: 4,920,083	7.3	69	0.34	*Ananas bracteatus* clone 49764a microsatellite sequence	100/99	1e -26
					Ananas comosus postacrosomal sheath WW domain-binding protein (LOC1019711040) mRNA	100/97	6e -25
4710330	Scaffold 1169: 12,423	6.9	69	0.47	*Ananas comosus* receptor kinase-like protein Xa21 (LOC109704031) mRNA	100/100	3e -28
					Ananas comosus receptor kinase-like protein Xa21 (LOC109710323) mRNA	100/97	6e -25
					Ananas comosus LRR receptor-like serine/threonine-protein kinase EFR (LOC109710348) mRNA	100/96	3e -23
					Ananas comosus LRR receptor-like serine/threonine-protein kinase GOS1 (LOC109704033) mRNA	100/96	3e -23
4724753	Scaffold 1089: 42,316	6.8	69	0.36	*Ananas comosus* receptor kinase-like protein Xa21 (LOC109703920) mRNA.	100/96	3e -23
					Ananas comosus LRR receptor-like serine/threonine-protein kinase GSO1 (LOC109704033) mRNA	100/93	6e -20
					Ananas comosus receptor kinase-like protein Xa21 (LOC109703919) mRNA.	100/91	3e -18
					Ananas comosus receptor kinase-like protein Xa21 (LOC109710578) mRNA	100/90	1e -16
					Ananas comosus putative receptor-like protein kinase At3g47110 (LOC109710347) mRNA	100/90	1e -16

					Ananas comosus LRR receptor-like serine/threonine-protein kinase GSO1 (LOC109710346) mRNA	100/90	1e -16
					Ananas comosus receptor kinase-like protein Xa21 (LOC109710331) mRNA	95/91	1e -16
					Ananas comosus receptor kinase-like protein Xa21 (LOC109710330) mRNA	100/90	1e -16
					Ananas comosus LRR receptor-like serine/threonine protein kinase EFR (LOC109710348) mRNA	100/88	6e -15
					Ananas comosus receptor kinase-like protein Xa21 (LOC109704031) mRNA	100/88	6e -15
					Ananas comosus receptor kinase-like protein Xa21 (LOC109710811) mRNA. Transcript variants X1-2	100/87	3e -13
					Ananas comosus probable LRR receptor-like serine/threonine protein kinase At3g47570 (LOC109710816)	79/91	4e -12
					Ananas comosus receptor kinase-like protein Xa21 (LOC109710128) mRNA	88/89	4e -12
					Ananas comosus receptor kinase-like protein Xa21 (LOC109710334) mRNA	100/86	1e -11
					Ananas comosus receptor kinase-like protein Xa21 (LOC109704034) mRNA	100/86	1e -11
100067858	LG05: 4,886,755	6.8	69	0.49	*Ananas comosus* probable Xyloglucan endotransglucosylase/ hydrolase (LOC10970994) mRNA	100/99	1e -26
4719439	NP	6.5	40	0.50	*Ananas comosus* uncharacterised protein (LOC109710749) Transcript variants X2, X3	95/100	5e -11
4717643	LG05: 5,037,139	5.7	69	0.31	*Ananas comosus* transcription factor bHLH 128-like (LOC109710976) Transcript variants X1-3	81/98	2e -19

*Marker #4720783 is positioned on a Hypothetical protein. This is in close proximity to a α/ β hydrolase, β-glucosidase and Xyloglucan endotransglucosylase/ hydrolase.

Table 2. Putative genes in LD with SNP #100029071. Only marker pairs with an R^2 >0.5 are shown. Positional matching of the markers does not imply the genes listed match the marker sequence.

Marker 1	Position 1	Marker 2	Position 2	Gene	Distance (kb)	R^2	D′
#100029071	LG05: 4,708,927	#4715294 On Aco004620.1	LG05: 4,514,730	60S ribosomal L7a	1,941	0.8	0.9
		#4722846 On Aco004602.1	LG05: 4,351,819	Microtubule-associated protein 65-8	357	0.7	0.9
		#4715322 On Aco004551.1	LG05: 3,922,000	TSL-kinase interacting protein 1	787	0.6	0.9
		#4726821 Flanks Aco004595.1	LG05: 4,278,331	Terpenoid cyclase	431	0.6	0.9
		#100040141 On Aco004607.1	LG05: 4,390,401	Hypothetical	319	0.6	0.9
		#4716089 Flanks Aco004552.1	LG05: 3,622,796	Glutathione S-transferase	1,086	0.6	0.8
		#4715878 On Aco004590.1	LG05: 3,766,127	Glycosyl hydrolase	943	0.6	0.8
		#4715706 On Aco004571.1	LG05: 3,941,586	Inositol-1,4,5-triphosphate 5-phosphatase	767	0.6	0.9
		#100046716 On Aco004495.1	LG05: 4,230,539	CBS domain-containing	478	0.6	0.8
		#4711595 On Aco004571.1	LG05: 4,108,202	GTP cyclohydrolase	601	0.5	0.8
		#4718072 On Aco004495.1	LG05: 3,584,460	Mitogen-activated kinase	1,124	0.5	0.8

Table 3. Genes co-expressed with Glycosyl hydrolase (Aco004528) in the roots of 'Smooth Cayenne' (The Pineapple Genomics Database of Zhang and Ming, (2018); Xu et al, (2018); http://pineapple/angiosperms.org/pineapple/html/index.html).

Gene	FPKM	Position	Function	Reference
metallothionein 3 (Aco009983.1)	1495	Scaffold 15: 1,977,111; LG10: 1,977,111	Metal chelators involved in metal homeostasis and oxidative stress protection. Can be up-regulated in response to pathogens.	Miles et al, (2011)
Cysteine inhibitor (Aco021907.1)	657	Scaffold 166: 122,955; LG13: 4,569,463	Cysteine-rich proteins produced by pathogens can be pathogen-associated molecular patterns (PAMPs) in some plants that induce a hypersensitive cell death response.	Nie et al, (2019)
NADH dehydrogenase ubiquinone 1 beta sub-complex subunit 3-B (Aco003402.1)	170	Scaffold 5: 1743533; LG17: 2626718	Also called NADH-quinone oxidoreductase, is a major source of ROS.	Kussmaul and Hirst, (2006)
Protein of unknown function DUF1664 (Aco000434.1)	131	NA	NA	
DNA-directed RNA polymerase subunit 10-like protein (Aco003288.1)	106	Scaffold 5: 869,215; LG17: 1,752,400	RNA polymerases are involved in RNA silencing and are associated with resistance to viruses and invading iRNAs. Considered part of the genome-wide systemic-acquired resistance response.	Willmann et al, (2011)
ferredoxin 3 (Aco022117.1)	97	Scaffold 188: 36,855; LG05: 8,638,708	Ferredoxin is a cofactor in biosynthesis of fatty acids used in the JA pathway.	Kachroo et al, (2003)
Endo-1,3-1,4-beta-D-glucanase (Aco006291.1)	94	Scaffold 17: 380,939; LG14: 3,514,104	β-glucanases are parthenogenesis-related proteins that hydrolyze the β-glucans in most fungal cell walls.	Singh et al, (2014)

PSY1 captures key pineapple plant physiological process

ICT International Pty Ltd, 211 Mann St., Armidale NSW 2350

The pineapple plant assimilates CO_2 via the crassulacean acid metabolism (CAM) pathway – an evolutionary adaption to arid environments. According to literature, moisture inside CAM plants is conserved when stomata remain shut during the day. For pineapple, the stomata are closed after sunrise until mid-afternoon (Bartholomew and Kadzimin, 1997).

A recent investigation using ICT International's Psychrometer (PSY1) revealed the dynamics of plant water relation in pineapple at high temporal resolution (Figure 1). The measurement was done using a representative pineapple plant grown in the field in South Sumatra, Indonesia (Figure 2). Although not replicated, the preliminary study describes the physiological behaviour of pineapple elucidating CAM mechanism. Water potential measurements were highest in the morning (when the stomata are closed) and lowest close to sunset. Furthermore, night-time water-potential values sit somewhere in the middle potentially suggesting transpiration when the environmental evaporative demand is low.

Editor (DPB) comment: The PSY 1 should be a valuable tool for anyone studying the ecophysiology of pineapple in natural environments.

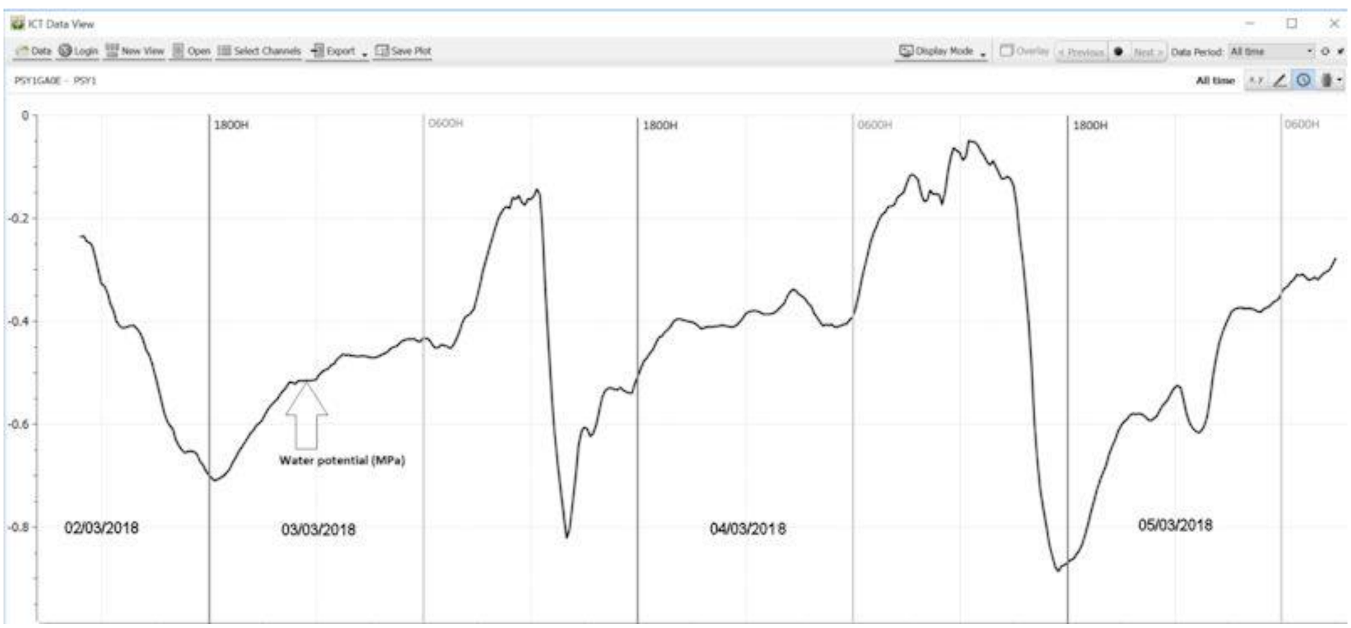

Figure 1. Diurnal pattern of plant leaf water potential (MPa) of a pineapple leaf.

Figure 2. Pineapple plant with the insulated psychrometer (PSY1) installed in the leaf (Inset: PSY1 instrument).

News from Costa Rica

Data required for simulation modeling of pineapple production with DSSAT

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The objective of modeling an agricultural crop is to "simulate production over time and space for different purposes" (Jones et al., 2003). Much time and money has been invested in modeling the major annual crops produced in more temperate environments while considerably less time and money has been invested in the modeling of some of the important tropical crops have been neglected.

The effects of temperature on plant growth were observed centuries ago and the summing of temperature information and relating it to plant phenology dates to the 18th century (Wang, 1960). Modeling of pineapple is a rare activity that apparently began at the Hawaiian Pineapple Company (later named Dole Pineapple Co.) plantation on Lanai Island, Hawaii (Medcalf, 1949). Many years later a heat unit model for the prediction of fruit development of 'Smooth Cayenne' was published (Fleisch & Bartholomew, 1987) and not long afterward the more comprehensive 'Smooth Cayenne' model ALOHA-Pineapple (the Model) was developed (Zhang & Bartholomew, 1993; Malezieux et al., 1994; Zhang, et al. 1997). The Model was incorporated into early versions of the Decision Support System for Agrotechnology Transfer software (DSSAT; version 4.7.5 is available at no cost at https://dssat.net) (Hoogenboom et al., 2019; Jones et al., 2003). The pineapple crop model SiMPiNA was developed recently in Reunion Island (Dorey, et al., 2015) but it is both site and cultivar ('Queen') specific so likely would need major revisions to be of use with other cultivars and locations.

Lack of interest in pineapple crop modeling left the Model unsupported until 2017 when Dr. Rey Umali and colleagues at Dole Philippines (now owned by ITOCHU Corp.) contracted with the Dr. G. Hoogenboom and colleagues at the University of Florida to update the Model so it runs in DSSAT 4.7. Workshops on the functions of DSSAT and the Model have been made to two pineapple companies and DSSAT workshops are conducted annually. The DSSAT 2019 International Training Program took place on May 20-25, 2019 at The University of Georgia, Griffin, Georgia, USA and the next one is scheduled for May 20-23, 2020 at the same location. The 2019 workshop covered the topics: Assessing Crop Production, Water and Nutrient Management, Climatic Risk, and Environmental Sustainability with Simulation Models.

Weaknesses in the Model, especially loss of the supporting data sets originally used to test and validate the model in multiple environments, have been identified. Pineapple culture also has changed dramatically since the fresh pineapple industry expanded rapidly after 'MD-2' was introduced into world markets. Some of the weaknesses of the Model are identified below. We have made an attempt to identify some of practices that must be included in the Model and also try to flag the most significant weaknesses. We believe that pineapple farms at all levels would benefit from having a robust model that would help manage current culture processes but also identify future problems, e.g., global warming, would be difficult for any company or country to prepare for. Despite its international importance, pineapple remains among the tropical orphans when it comes to research that would benefit all growers with little impact on their company markets.

DSSAT is a great modeling program for crops that have enough information to project growth and production. The DSSAT package includes simulation models for more than 42 different crops and the information for each crop includes three different types of variables that affect growth and productivity. These types are variables inherent to the crop species or cultivar, variables that can be modified by management, and finally those corresponding to the environment and the specific climate of each season or production campaign.

On the other hand, DSSAT also has different tools, applications and simulation options that offer very diverse and useful modeling and prediction possibilities that are also based on the information of the variables mentioned above.

In order for DSSAT to model the growth of a crop, the above variables must be organized or parameterized in a module. For example, the modules for rice, corn, soybean and alfalfa contain the variables organized so that they can be useful to DSSAT. For these deeply researched crops there is a lot of information in their respective modules captured with enough scientific and technical rigor so that DSSAT can project or predict crop growth and productivity with adequate precision. These annual crops have very specific sowing windows and harvest dates (not sown all year) so even though they are sown in large areas, the number of trials required to improve or increase the response variables is relatively small. In contrast, even on relatively small farms, fields (minimum management units or MMU) of pineapple are planted almost every week of the year, its crop life cycle is never less than 15 months (soil preparation/planting to harvest) and it can be a perennial crop in many areas where it is grown. Since each week of the year will expose the crop to a permutation for the other variables, it greatly expands the number of different results required to build an adequate database. In addition, quality requirements for this fresh fruit are very different from those for processed pineapple, so there are a large number of technical considerations in the last weeks of fruit development that must be taken into account for optimum crop yield.

The module that DSSAT uses to project the growth of a pineapple crop is ALOHA-Pineapple. The concepts for that module were developed for the Smooth Cayenne cultivar, which is still grown, but primarily for processing. Dietary preferences have greatly expanded the demand for fresh pineapple, which because of continuous market demand and fruit fragility has greatly increased the importance of production scheduling and has introduced new cultivars, the predominant fresh fruit cultivar being 'MD-2'.

In order for DSSAT to be useful in modeling the multiplicity of pineapple cultivars growing in a wide range of environments, information capture must be done for each cultivar over the range of environments where it is grown. And if the data sets come from the diverse agroecosystems where pineapple is grown the consolidation of the information will provide the datasets required for the improvement of the Model.

The following sections are the result of a DSSAT review focused on the Model

1. Minimum data set concept:

The concept of a minimum data set is what makes it possible to run various models in DSSAT. This dataset corresponds to the minimum variables required to be able to project the growth of a crop and includes the following: Required weather data including latitude and longitude of the weather station, daily solar radiation (MJ/m^2-day), maximum and minimum daily air temperature (ºC), and daily total rainfall (mm). The minimum data set also includes desired soils data relevant to pineapple and management data for pineapple. If the Model is to be useful from a practical point of view, the management data must be consistent with the main farm practices. We consider the following points as the most relevant to take into account in the minimum data set for a pineapple model, since physiology and management definitely involve processes very different from the typical model in DSSAT.

a. Cultivar
While several pineapple cultivars are grown on a commercial scale, only ‘Smooth Cayenne’ and ‘MD-2’ are of major importance. The Model was developed using data for ‘Smooth Cayenne’ so basic data for that cultivar are already in the Model. Market requirements, fruit fragility, cultivated surface and importance in the world markets make it clear that any initial effort would focus on ‘MD-2’. It is expected that focusing on ‘MD-2’ will speed development of the Model and once the Model is functional for one cultivar, it will be relatively easy to incorporate other cultivars that are grown on a much smaller scale.

b. Initial weight of planting material
Currently Model initiation begins with the entry of data on the planting material dry weight per hectare. To be consistent with farm practice, the seed piece (propagule) average fresh weight needs to be used to establishes the initial plant mass. Pineapple planting material consists of crowns, slips and suckers, or more than one of those, and is planted practically every week on commercial farms. Only one type of planting material of uniform size would be planted in a minimum management unit (MMU, typically a block twice the width of the farm sprayer boom and of variable length)

The fresh seed weight varies with the type of seed and generally seed weight increases from crowns to suckers and they have different characteristics (see Botany and Physiology in Py et al., 1987). Seed size within a field must be uniform so all plants have nearly equal space in which to grow. Variation between MMUs is fairly common because it is determined by the type and size of propagule available at the time of planting. Within a specific environment, the greater the initial weight of the seed pieces, the shorter the days from planting to forcing.

c. Planting date
The planting date, simple as it seems, is one of the most complex variables that must be part of the minimum data set to improve the current version of the Model. The climate varies with the season and location of the farm in the tropics and subtropics so the plants in each MMU are almost certain to be exposed to a different and gradually changing environment. For more details, see “simulating potential production and Sensitivity analysis tool” below.

d. Density (bed, row and plant spacing)
The density in the pineapple crop is different from that of many modeled crops because the typical layout is two-row beds with an interspace between two such beds to provide access for field workers for weed control and harvesting. Density then is determined by distances from bed center to bed center, and between plants in the row. Planting density may be lower in low-elevation equatorial regions because in such environments pineapple has a lower harvest index (see ahead) than it does where the average temperature is lower. Typical planting densities range from about 80,000 to 120,000/ha for cultivars with small fruit, e.g., ‘Queen’, to 50,000 to 80,000 for cultivars with large fruit, e.g., ‘MD-2’and ‘Smooth Cayenne’.

e. Monitoring growth until forcing
Once planted, all subsequent plant measurements made before the harvest should show the average weight of an individual plant. It is assumed that any grower interested in

modeling pineapple production will take advantage of the opportunity to force reproductive development rather than relying on the random natural induction provided by nature.

We think it is very important that the software be equipped with some field information capture screens, similar in concept to the Pineapple Crop Log (Sanford, 1962). An alternative would be to use a stationary monitoring plot for each Plantation Development Group (PDG; group of MMU over which the same technological package is applied) on the farm. This stationary monitoring plot (it is a MMU within the PDG) should always be sampled (weight, and other growth variables, leaf analysis, pests and diseases) and becomes part of the minimum data set for modeling purposes.

In order for DSSAT to model growth and make projections, the information required to monitor average plant fresh weight must be captured. The pineapple producer has to decide when to induce fruiting. This means that the model must facilitate the decision to induce fruiting at the appropriate time to maximize marketable kg/ha of fruit of acceptable quality and the best organization within the area for the best operation of the fruit harvest.

2) Soil Data Inputs and Utilities

The model must allow for the entry of soil analysis results for Al, Mg, K and Ca because they are required to calculate important fertilization properties in variable charge soils (typical of the tropics). Also, if not currently available, DSSAT must accept foliar

3) Simulating Water-Limited Production and Effects of Extreme Weather

It is unlikely that simulation of water-limited production of pineapple will be a component of the Model any time soon. (Carr, 2012) noted that as a result of its CAM metabolism pineapple differs from most other commercial crops and (Ekern, 1965) showed that evapotranspiration was lower at midday for large and older plants with a greater LAI than for much younger plants with a smaller LAI. (Carr, 2012) thought it remarkable that given the importance of pineapple as an internationally traded commodity there were few reliable publications quantifying where "irrigation of pineapple is likely to be worthwhile, how it is best practiced and the benefits that can be obtained."

Many soils under pineapple cultivation have a high clay content, which gives them a high crop moisture retention capacity that together with pineapple's CAM metabolism reduces the adverse effect of a few dry months. Some areas of Africa, Colombia and Mexico have lighter soils and experience serious problems with the growth and productivity of the pineapple crop due to prolonged periods of water shortage. Where DSSAT can model differences in soil texture and water holding capacity in water-limited environments, it could allocate limited irrigation water to MMUs that would have the greatest impact on farm income if plants were severely stressed, e.g., MMUs where fruits are within a few weeks of harvest.

As noted previously, weather data is required for DSSAT and crop models to function. While weather data is required to simulate pineapple growth, it could also be important when weather extremes can cause significant crop losses. For example, quantify sunburn of the fruit and injury of fruit crowns. Sunburn of fruit can occur at and above 22 Mj / m^2 in Costa Rica. Some farms place shade cloth over plants most susceptible to sunburn injury. In contrast, Hawaii crown and fruit sunburn percentages can be small to nonexistent at 27 Mj / m^2 if air temperature is less than 30°C and the wind velocity is 5 m s^{-1} or greater. The wind rapidly transport heat away from tissues and structures by forced convection. The truth is that sun burn in the pineapple is important for the fruit, flower and leaves so it should be considered in the Model. Wind data must be collected in order to model energy balance calculations to predict the probability of sunburn.

4) Simulating basic growth processes – the importance of the harvest index

When considering basic growth processes within a given environment, one of the most important objectives is to know or find the harvest index for that environment. For the sake of clarity, the definition of the harvest index for pineapple is fresh weight of the fruit at the time of harvest / fresh weight of the plant at the time of forcing (Hepton, 2002). A good model would provide an opportunity to examine the effect of leaf area index (LAI) and Canopy Net Assimilation Rate (CNAR) on packable yield. It is possible that if the canopy is reduced by reducing the planting density or the plant size at the time of forcing, it might increase the CNAR. Reducing the LAI would reduce interplant competition and improve light distribution through the canopy. It is possible that there is an optimum LAI that would significantly improve the uniformity of fruit size and maturation. A good model also could make it possible to predict the interaction of CNAR and yield across seasons. Presently it is standard practice to select a planting density that remains the same throughout the year. Reasonably accurate simulation of these interacting effects would be inexpensive whereas studying these same interactions in field trials would be prohibitively expensive.

It is possible that the differences in harvest index discussed below are also due to differences in the CNAR of the plants grown in the two hypothetical environments. Because pineapple crops at typical planting densities have very high leaf area indices, perhaps 10 or more, planting density is one of the strategies that could be explored to improve CNAR and, as a possible result, improve fruit size uniformity and quality. However, it is important to find the optimal point, since lowering the density could increase CNAR at the expense of fruit yield that could greatly decrease profitability.

Finally, a curiosity currently in the Model is that a variable of productivity is defined as number of eyes per square meter. This concept could be a carryover from the CERES maize model, which was modified to simulate pineapple growth and yield. For corn, the components of yield are kernel size, kernel number per cob and cobs per unit area. Similarly, for pineapple, the components of yield are fruitlet size, fruitlet number per fruit and fruits per unit area. Fruit weight is determined primarily by fruitlet number (Py et al., 1987), thus fruitlet size is less variable than fruitlet numbers per fruit so the prime objective of management would be to produce the greatest number of fruits per unit area with the greatest possible number of fruitlets.

5) Simulating potential production

This simulation option would be used on pineapple farms to make comparisons of production with different types of seed (assumes more than one type of seed is available), weights of seed, dates of planting and dates of induction of flowering. The analysis would help to identify the type and weight of seed and date of sowing that can achieve the highest productivity at a given date of induction, for example, with a special interest where the induction date is close to the time of natural induction (NI). Such an analysis would identify which type and weight of seed would make it possible to force fields near the time of NI to minimize the adverse effects of precocious fruiting. Such an analysis would be an important addition if it could make comparisons of different plantation management practices (fertilization, plastic mulch, etc) that might influence the percentage of NI.

6) Sensitivity analysis tool

In geographical locations where there is considerable seasonal temperature variation it is reasonable to think that there could be seasonal differences in the harvest index.

Sensitivity analysis would be fundamental for comparing the effect of planting material characteristics, mainly seed fresh weight at planting, on days from planting to the target fresh weight for forced inductions. This could help the farmer to realize a priori the repercussions in costs and any other adverse effects of the use of one or another of the seed pieces available on the plantation through the different months of the year. The goal is to predict with confidence the ideal combination of seed weights and days to forcing to obtain the best possible yield.

Where growers manage fields to produce planting material, a mistake made by many is to consider areas set aside for seed production as remnant areas so they receive minimal management. Active management of plots set aside for seed production is as important as for those areas that are developing fruit. The problem is that habit and the lack of awareness tools like DSSAT prevents growers from having a wholistic view of the entire production system. For this same reason, the production of seeds and the production of ratoon crops require independent simulation modules in DSSAT.

7) Using DSSAT to Simulate Crop Rotations in Long-term

This is potentially one of the most valuable applications in DSSAT because in pineapple production it is one of the concepts most overlooked by producers. Producers waste valuable resources because they do not know, or at least do not consider, management of the cultivation cycle as an important operative goal.

Most crop models in DSSAT are for annual crops that operate with a relatively small planting window and practically all the tillable land is planted or handled in a very short time. Also, harvest date for annual crops is predictable but prediction could be irrelevant because some crops, for example cereal grains, can remain unharvested in the field for days after the crop has matured. Thus the application "Simulating Crop Rotations in Long-term", is best suited to annual crops that are sowed in very little time (a week for example), then the application simulates the rotation of crops for the entire surface of the farm. This situation is relatively easy to program because there are very few options due to the very defined and limited sowing window.

The pineapple farm is planted week by week, so the farm is managed by MMUs and each has a unique planting date and is just a fraction of the total tillable land area. This particular application of DSSAT would force entering MMU as if it were the entire farmed area (because it is by planting date). When the simulation applications is applied to pineapple, it would force the user to enter all MMUs to be able to model the entire farmed area. However, the application would only allow us to see the modeling of MMUs and not the whole farm, which is not the desired objective.

For this application to be functional for a pineapple farm with multiple MMUs, it should be possible to group them and present them all together, but that is currently not possible. The number of operational decisions that affect pineapple stages in each MMU is a peculiarity of the crop, so that long-term modeling becomes corrupt in a very short time.

Some operative decisions that can affect the duration of the cultivation schedule of one MMU are below. It is clear that it is necessary to endow this DSSAT application with a set of manual corrections / manual adjustment options by MMU:

1. Timing of induction of reproductive development (forcing) of pineapple is determined mainly by plant weight and the grower. Fruit harvest can occur five or more months later and can vary from the predicted date by a few weeks. It is necessary that the application project maturity (date of harvest) for some number of degree days. However, relatively short changes in the average air temperature can advance or delay the actual date of harvest by several days to a few weeks. If the field is to be managed for the production of a ratoon crop or planting material, the model must accept the manual entry of the actual date

of harvest so that simulation of the next stage in the life of a particular MMU can be modeled from the beginning of the stage.

2. Pineapple fruit is degreened to satisfy the market requirement that the fruit skin has at least some natural color. The date of degreening is determined by the actual average fruit °Brix, which fixes the date of harvest within a few days. However, the differences in days "could" not be very significant. Degreening could require a Model manual correction to account for natural/unanticipated changes in °Brix as a result of an atypical delay or advancement in that measurement.

3. After fruit harvest, a MMU can be managed to produce a second (ratoon) crop, seed, primarily suckers, or prepared for the next crop. The duration of each of those options differ greatly.

4. Operational efficiency in decision-making: this can be a big problem because it is not uncommon for producers to take a month or more to establish a plan for a recently harvested MMU. This undoubtedly will significantly affect the duration of the culture cycle (see more below).

5. Another condition that can affect the duration of the crop cycle is nutrition, which depends on an adequate and on-time supply of fertilizers. Another peculiarity of pineapple culture is that nutrition is predominantly through foliar fertilization on a schedule rather than being based on plant requirements. Any significant delay would slow growth and increase the days required to reach the target plant weight for forcing.

Due to the above, in order for DSSAT to simulate the long-term crop cycle, it must allow the entry of MMUs. A hypothetical farm with 300 net hectares planted to pineapple could have up to 1000 0.3 ha MMUs of which approximately 350 are planted annually. The other MMUs would be in one of the other stages of the cycle mentioned above. Considering all the management factors mentioned above that can affect the duration of the various stages, the question is how to model the MMUs so the stage of cultivation of each unit can be projected, perhaps up to the year 2025. While the hypothetical 0.3 – 0.6 ha is a typical MMU in Costa Rica, the topography of each farm will determine the size of the various MMUs on the farm. Since simulating pineapple production on a 300-ha farm with 1000 0.3 ha units with DSSAT would not be an unusual scenario, an example is illustrated below.

The indicator of operational efficiency will be the hectares available to plant each year and therefore the number of boxes of fruit to be harvested per year:

The scenarios are:

1. Plant crop with no and one ratoon harvests.
2. Soil preparation time.
3. Productivity of the seed production plots. The higher the productivity, the smaller the area devoted to seed production to meet the planting goal and the sooner the land can be replanted.
4. Harvest time: In most cases the harvest time is 15 days. However, in this case a block of time was added so the farmer can decide what to do with the harvested area.
5. Time from planting to forcing: In the following scenario, it is assumed that forcing was delayed because the fertilizers were not applied in a timely manner. As a result, the growth rate was lower, and it was necessary to delay forcing until the target plant weight was reached.

Table 1. First Crop or First and Ratoon Crop production model where annual projected harvest is 1,426,345 boxes/year for a farm of 300 net hectares. Area is in hectares and "%" is the fraction of the farm devoted to that operation (In the table, one plot is equivalent to one minimum management unit or MMU).

MODELO FIRST CROP AND RATOON CROP

Etapa	Duration in months	AREA	%
Soil preparation	4	**57**	19%
Planting (time planting one Plot)	0.5	**7**	2%
Planting development	8	**113**	38%
Harvest (time harvesting one Plot)	0.25	**4**	1%
Fruit development	5	**71**	24%
Development ratoon	8	**113**	38%
Development fruit ratoon	5	**71**	24%
Harvest ratoon (time harvesting one Pl	0.25	**4**	1%
Seedlings plots (where we ged seeds)	2.2	**31**	10%
Totals	33.2	**471**	157%

CURRENT FARM AREA (HA)	300		
		by Year (ha)	by Month (ha)
Month planting (equal forzing and harvest)			9.04
Total annual first crop		108.48	
Establishment of ratoon crops			9.04
Total annual harvest ratoon crops		78.35	
Productivity first crop (Ton / ha)	100	10848	
Productivity ratoon crop (Ton / ha)	80	6268	
TOTAL ANNUAL TONS		**17,116**	**1,426,345**
Rejection Ton / ha first crop	15	**1,627**	
Rejection Ton / ha ratoon crop	20	**1,567**	
Seedling productivity (seeds / plant / month)			0.3

Table 2. First Crop and Ratoon Crop model, 1,137,584 boxes for a farm of 300 net hectares, an additional month allocated for soil preparation, a drop in the productivity of the seedlings plots, loss of time after harvest and with one more month from planting to forcing.

MODELO FIRST CROP AND RATOON CROP

Etapa	Duration in months	AREA	%
Soil preparation	5	**71**	24%
Planting (time planting one Plot)	0.5	**7**	2%
Planting development	9	**128**	43%
Harvest (time harvesting one Plot)	1.25	**18**	6%
Fruit development	5	**71**	24%
Development ratoon	9	**128**	43%
Development fruit ratoon	5	**71**	24%
Harvest ratoon (time harvesting one Pl	1.25	**18**	6%
Seedlings plots (where we ged seeds)	2.7	**38**	13%
Totals	38.7	**549**	183%

CURRENT FARM AREA (HA)	300		
		by Year (ha)	by Month (ha)
Month planting (equal forzing and harvest)			7.76
Total annual first crop		93.08	
Establishment of ratoon crops			7.76
Total annual harvest ratoon crops		54.29	
Productivity first crop (Ton / ha)	100	9308	
Productivity ratoon crop (Ton / ha)	80	4344	
TOTAL ANNUAL TONS		**13,651**	**1,137,584**
Rejection Ton / ha first crop	15	**1,396**	
Rejection Ton / ha ratoon crop	20	**1,086**	
Seedling productivity (seeds / plant / month)			0.2

8) Summary:

Many of the DSSAT models are well suited for extensive crops with a small sowing window while its utility is limited for the MMU structure of pineapple management that involves planting a unit about every week. It seems that for DSSAT to be functional for the cultivation of pineapple, first the model must be fed with enough data to be able to model the different scenarios that a given producer could have. And one doubt is how far to go with DSSAT in pineapple. For example, if the software will be limited to making specific projections of a certain number of variables for a single MMU, it will be much less useful than if it can be taken to a higher level and allow the producer to project what will happen to all the MMUs on his farm in a consolidated manner (all MMUs projected). The two scenarios are very different and the latter whole farm capability is a much more valuable management tool.

The DSSAT Minimum Data Set is clear, general climate data are clear and soil data are clear, but what are the crop data, or the growth variables required in any trial done to add cultivars and improve ALOHA-Pineapple, Data collection for the SIMPIÑA model for 'Queen' (Dorey et al., 2015) included monthly measurements of dry and fresh weights of leaves, roots, stems, peduncles, inflorescences, fruits, and crowns and the number of fruitlets per fruit for eight pineapple plants in each replicate. It is expected that additional data will be required for ALOHA-Pineapple to conform to the stricter and more flexible DSSAT software. Below are many of the crop variables that we believe must be evaluated to be certain that the crop data set is adequate for modeling pineapple.

For example:

1. Type of seed: (slip, sucker, ground sucker, hapa, crown).
2. Seed pruned or unpruned (This deserves its own category; No publications on this treatment were found).
3. Fresh weight of seed to be planted: (task to define universal table of ranges of seed weights).
4. Number of leaves at sowing: count per plant (sampling of number of leaves at the time of planting).
5. Leaf area index (LAI): (task to define universal methodology to quantify the leaf emission rate and measure LAI remotely).
6. Sampling frequency (no less than monthly but may reduce frequency based on growth rate and stage and the variables to be measured).
7. Fresh weights of each plant and, based on objectives, dry weights of components for each sampling event.
8. D leaf length, width and weight at each sampling event.
10. Fertilizer requirements (based on LAI or canopy color, or both) and foliar nutrient application.
11. Impact of water stress on the growth responses of the main commercial cultivars.

After the forcing the data required could be:

10. Days from forcing to 1 cm open heart (visible opening of 1.0 cm).
11. Days from forcing to anthesis of flowers in the first row of fruitlets and number of fruitlets per fruit.
12. Date 12.5º Brix reached (degreening day).
13. Task to define a table of translucency or an internal color chart of the fruit and reserve the term translucency for an undesirable quality characteristic of the fruit (either because it came out

prematurely, or because it came out by natural ripening of the fruit, but equally undesirable because it would be a non-exportable fruit by age or internal condition).

14. Harvest day after the day of degreening.

One way to get the minimum data set for ALOHA-Pineapple could be get the information through modifications of the sampling system currently used on well managed pineapple farms. However, the sampling methodology (what we need to measure, when we need to measure and how we have to measure) must be uniform. Grower cooperation would greatly speed collection of the minimum data set, but it would require many compromises to guarantee the quality of the information collected by the farms or companies involved in the work. The other way (maybe the most confident), is from trials, although the same standardization of criteria are necessary. Today farms in Costa Rica in general terms are getting or looking for the same information in the field in order to make decisions. However, when you speak with any pineapple producer or technician about a specific methodology to get information, variation in criteria for the same process is very high between producers and technicians because of many conceptual or methodological errors in their processes.

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News from Cuba

Introduction and diversification of new pineapple cultvars in Ceballos Agroindustrial Enterprise. Cuba.

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The Cuban production of pineapple is based, almost exclusively on 'Red Spanish', which is vigorous and well adapted to cultivation, but has thorns on the leaves, a low yield potential and the fruits have deep eyes and a barrel form that reduces the use of its pulp. In 2009 the hybrid ´MD-2´was introduced in Pineapple Production Unit (UEB) of the Ceballos Agroindustrial Enterprise, which requires excellent agro-technology because of its susceptibility to stress, natural flower induction and fungal diseases (Bartholomew, 2009).

Since that year, fresh fruits of 'MD-2' have been produced and marketed to different European markets. Cuba has great genetic diversity such as different ecotypes of Red Spanish, Cabezona (triploids), Smooth Cayenne Serrana and the White Pineapple or Pineapple of Cuba (Isidrón et al., 2009) in the hands of the peasants. Limitations of those cultivars make it necessary to introduce more productive cultivars to the country and thus be able to evaluate their behaviour under Cuban soil and climatic conditions. Superior cultivars are introduced gradually to enterprise and the peasant sector to diversify the varieties. These will join already adapted cultivars that are grown to supply family consumption and small national marketing.

In searching for new cultivars, two working strategies have been established: 1- acquisitions of agamic propagules from others countries with recognized reputation in pineapple production and 2- introduction from micro-propagation techniques (vitroplants, Figure 1). The second will be acquired in the Technological Scaling and Transfers Laboratory of the Bioplant Center in Ciego de Ávila, University, Cuba, which has vast experience in pineapple vitroplantas production and marketing. The company also intended to gradually introduce *in vitro* such cultivars as Smooth Cayena, Perola, Vitoria and others of interest in the world market.

Figure 1. Synthesized diagram of the 'MD-2' pineapple vitroplants production. Photo of L. Nápoles)

At present, 'MD-2'and the Smooth Cayenne clone 'Champaka' produced *in vitro* from existing material in the germplasm bank of the Bioplant Center are already found in the experimental areas of the UEB. In transfer vitroplants to field conditions, the aim is to link the experiences that UEB has achieved in agro-ecological management with the adaptation of some results realized by Mexico in the Intensive Pineapple Production System Under Protected Environment (Uriza-Avila *et al*., 2018), which use plastic mulch in soil and shade mesh (Black Saran 50%) with excellent production levels.

Figure 2 Pineapple vitroplants (Champaka and MD-2) introduced in field production. Photos of R. Rodríguez)

In the agro-technical process established for the field introduction of vitroplants, bio-products produced in Cuban laboratories are used, such as: *Trichodermas, Basillum* (LBT 13), *Bauberia*, Micofert (mycorrhizae), efficient Micro-organisms, Analogue of Brassinosteroids (Biobras-16) and earthworm humus (solids and liquids), all of them in a well-structured application program according to the requirements and the development phase of crop.

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News from Dominicum Republic

X International Pineapple Symposium

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The X edition of the International Pineapple Symposium will be held between 22 and 26 of April 2020 in the heart of the Caribbean, Punta Cana (Dominican Republic). The Symposium will happen at a time when the pineapple sector is facing big challenges in the whole value chain considering the dynamic environment of new actors growing and commercializing pineapples.

The symposium will be focused on the phytosanitary and post harvest control challenges in order to preserve the global market for pineapples. Each challenge or opportunity will be addressed with an economic perspective to provide the participants with a realistic insight of the feasibility or incidence of the situations or proposals. Being biodiversity protection and management a concern for the agriculture of the 21st Century, the topic will also have its space to listen the most relevant experience of biodiversity in pineapple sector.

The X Symposium will close with a tour around the plots and packing house of a new and promising project of pineapple, located in Monte Plata. Of course, the Symposium is a great opportunity to create and strengthen networking in pineapples business worldwide. Finally, why not to mention the paradise around the convention center, the first tourism destination in the Caribbean with top beaches.

News from France

In memorium: Pierre Martin-Prével,

Jean-Pierre Gaillard (provided by Guy Martin-Préve) with additional comments by D.P. Bartholomew

CIRAD (retired)CIRAD alumnus, Pierre Martin-Prével, passed away on December 24, 2018, at the age of 89.
He was born in Versailles in 1929, in a family of four boys. He obtained his education in Versailles and earned an agricultural engineer diploma at "Ginette" in Versailles. Later he obtained a postgraduate degree in plant physiology at ORSTOM. He conducted research on the effects of potassium, nitrogen and other nutrient elementS on anomalies in the quality of the banana, especially the yellow pulp, as part of a doctorate thesis. However, he never presented his work to a jury.

Pierre was recruited by the Institute of Colonial Fruits and Citrus (Ifac) and was assigned to the central station of the institute in Foulaya, Guinea as a researcher in charge of research on the nutrition of tropical fruit species and in particular of banana. After a stay of two and a half years, he was assigned to the Ifac plant analysis laboratory in Nogent-sur-Marne from 1957 to 1964. After analytical research there he joined the experimental field for a period of two years on the Azaguié station in Côte d'Ivoire where he mainly studied the mineral deficiencies of bananas on boxes lisymétriques.

Pierre returned to headquarters of the Fruit Research Institute and citrus fruits (Irfa) in Gabrielle, France in 1967 as director of the plant physiology and laboratory service analysis of Nogent (avenue de la Belle) with the support of researchers Jean Marchal, Jean-Joseph Lacoeuilhe and Michel Folliot. From 1967 to 1975, before his assignment to the Gerdat in Montpellier, Pierre was particularly fascinated by the automation of certain mineral analyzes by adapting "Technicon" equipment (used at the time only in medical settings). For ten-years he organized and supervised most of the research conducted overseas on foliar diagnosis and nutrition of fruit species such as banana, pineapple, clementine, avocado, mango, cashew and papaya.

If Pierre Martin-Prével was well known in the scientific community of plant nutritionists, he was equally well known in the world of symphonic music and choral singing. Wherever he resided in France, he created and directed a choir especially in the context of the movement "A Coeur Joie" in Paris, Versailles, Montpellier and finally Rochefort. In his capacity as chief of heart, he orchestrated numerous lyrical events including the Palais des Congrès at Porte Maillot in Paris. He now rests at Chesnay, France.

Additional comments by D.P. Bartholomew.

Of particular interest to the international pineapple community, Pierre leaves us with a considerable body of research on pineapple nutrition. He was the senior or co-author of at least 20 papers on pineapple nutrition. In 1992 Pierre and several colleagues from CIRAD attended the 1st International Pineapple Symposium (IPS) in Honolulu, Hawaii in 1992. At that IPS Pierre presented the paper *Network research on pineapple in and with the CIRAD-FLHOR* (1993. *Acta Horticulturae* 334:467-471). Consistent with his presentation on networking, Pierre strongly encouraged the symposium organizing committee to establish a pineapple working group within the International Society for Horticultural Science (ISHS). The ISHS Workgroup Pineapple was formed in 1993 and since then has lead the organization of eight additional IPS, the most recent in Cuba in 2017. It also prompted me to start Pineapple News and for the last 26 years Workgroup Pineapple and Pineapple News has strived to keep that community connected.

News from Mexico

Humid Tropic Collection: The Mexican pineapple

Advances and challenges in innovation management

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The book, in Spanish, is available to download in 7 MB and 15 MB versions at:
http://ciestaam.edu.mx/publicaciones2018/libros/pinia-mexicana-frente-al-reto-de-la-innovacion.pdf

and for dissemination on mobile devices at: http://ciestaam.edu.mx/libro/la-pinia-mexicana/

About the book

In the framework of the technical assistance model for tropical crops, called Agencies for the Management of Innovation for the Development of Suppliers (AGI-DP), the CIESTAAM instituted the Humid Tropic Collection to publish the analysis of the global and national context of each of the crops served, as well as the findings and lessons learned from the AGI-DP experience. However, given the important technological advances and commercial activities achieved by the Mexican pineapple in recent years, in this book of the series was decided to put more emphasis on recent innovations that explain these advances.

The book begins with a historical review of the cultivation of pineapple, an analysis of its context international and national and the botanical characteristics of the plant. Later, and described in detail are the characteristics and fundamentals of technology for intensive pineapple production in a protected environment, mainly for the production of pineapple 'MD-2' destined for export, as well as in the pre and
post-harvest associated with it. Finally, the agronomic contributions are highlighted, and environmental and commercial issues and challenges inherent to these innovations are discussed.

The construction of this work required a strategic alliance between the Group of research Piña-INIFAP of the Papaloapan River basin and the group of CIESTAAM researchers, members of the Technical Unit in Management of the Innovations, which allowed important synergies and personal learning and institutional.

In short, this book recounts the important innovation process of the production and marketing of pineapple in Bajo Papaloapan, the main area production in Mexico, in order to be a tool for analysis and technical support to producers, advisers, suppliers and services, agroindustries, marketers and officials involved in such an important agricultural activity.

News from Sri Lanka

Sri Lanka pineapple improvement program

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INTRODUCTION

The pineapple (*Ananas comosus* var. *comosus*) belongs to the family Bromeliaceae and is the third most important tropical fruit crop after banana and citrus in terms of worldwide production (Rohrbach *et al.*, 2003). Pineapple is cultivated mainly for fresh or canned consumption and to produce juice which is the only source of bromelain, used in the pharmaceutical market. Pineapple fresh fruit and making processed pineapple products are important horticultural industry in countries with tropical climates (Moyle, 2004). Mauritius ('Queen') and Kew ('Smooth Cayenne') are the major pineapple varieties grown in Sri Lanka. Kew is not very popular as a fresh fruit due to low Brix value, high acidity, high juice content, hard in texture and pale yellow flesh color, hence it is preferred for fruit processing. But Kew has some desirable characters such as barrel shape, broad shallow eyes and spineless leaves except at the tip. Mauritius is grown here to cater to the local market as well as for the export market. It has high demand as fresh fruit due to the high Brix value, less acidity, high Sweetness Index, soft texture, moderate juice content and golden yellow flesh color. However, Mauritius also has some undesirable characters such as conical shape fruit and spiny leaves.

Hybridization between Queen and Cayenne is expected to generate superior varieties having desirable characters required for both fresh fruit and processing (Hadiati *et al.*, 2011). Hence, hybridization program was initiated at Regional Agricultural Research and Development Center, Makandura, Department of Agriculture, Sri Lanka using Kew and Mauritius as the parents in the year 1995 (Fernando and Somadasa, 1997). From the hybridization program at Regional Agricultural Research and Development Center, Makandura, four promising hybrid lines named Hybrid 1 (H1), Hybrid 2 (H2), Hybrid 3 (H3) and Hybrid 4 (H4). Among them, H3 and H4) were better in terms of fruit shape (barrel), broad eyes, color of flesh (dark yellow) and with spineless leaves (except only at the tip). Characters such as barrel fruit shape with broad shallow eyes are more beneficial than characters like conical shape fruit with projected eyes. This is because considerable amount of flesh is wasted when peeling of conical shape fruit with projected eyes. One of the pineapple ideotypes is the spineless leaves. It is cost-effective to farmers to cultivate, manage and harvest their crop. But Fruit quality of H3 and H4 hybrids was not at the acceptable level (low Brix value, low Sweetness Index, hard texture and high juice content) and should be further improved. Hence, in the present study H3 and H4 hybrids were backcrossed with Mauritius mainly to improve the Mauritius excellent fruit quality characters.

MATERIALS AND METHODS

The study was carried out at Regional Agriculture Research and Development Center, Makandura, starting from 2013/14 Maha season to 2016 Yala season. Hybrid3 (H3) & Hybrid4 (H4) were back crossed with Mauritius and H3 x Mauritius back cross produced 14 offspring and H4 x Mauritius back cross produced 21 offspring. Fruit characteristics, fruit quality parameters and leaf margin type of offspring derived from both backcrosses were evaluated.

RESULT AND DISCUSSION

Fruit characteristics

1.Fruit weight (g)

The weight of fruits produced by offspring derived from H3 x Mauritius and H4 x Mauritius backcrosses varied from 632.3 g to 1400.0 g and 560.0 g to 4200.0 g respectively. Most suitable pineapple fruit weight for both purposes (fresh fruit and processing) is 1500 g – 2000 g weight group. The smaller fruited varieties (> 800 g) may be suited only for fresh fruit while the others can be utilized both for canning and fresh consumption. Six offspring derived from H4 x Mauritius backcross produced preferred fruit weight (1500g - 2000g) for canning and fresh consumption. All the offspring obtained from H3 x Mauritius backcross produced fruits lower than 1500 g weight.

2. Crown / Fruit Ratio (%)

The crown of pineapple fruit must be single and crown / fruit ratio should be 50 – 150%. (UNICE standards for marketing and commercial quality control of pineapples). Thirteen offspring obtained from H3 x Mauritius backcross had single crowns and only 1 offspring produced fascinated crown. Out of fourteen offspring derived from H3 x Mauritius back cross, eleven showed 50 – 150 % crown / fruit ratio. Twenty offspring obtained from H4 x Mauritius backcross had single crown and one offspring produced crownless fruit. Eighteen offspring derived from H4 x Mauritius backcross showed 50 – 150 % of crown / fruit ratio.

3. Fruit shape

Four offspring derived from H3 x Mauritius backcross and 10 offspring obtained from H4 x Mauritius backcross produced cylindrical or barrel shape fruits. Fruit length and girth mainly affect on fruit shape. The most suitable shape is cylindrical or barrel mainly for fruit processing because even removal of fruit skin from top to bottom with minimum wastage by mechanical peelers. The conical and round shaped fruits are not acceptable for mechanical peeling because it cut too much at the base or it may not remove the skin at the tapered end.

4. Core diameter (cm)

Core diameter is also an important fruit character. The core diameter of fruits produced by offspring derived from H3 x Mauritius and H4 x Mauritius backcrosses varied from 2.0 cm to 5.2 cm and 2.0 cm to 4.5 cm respectively. Higher width of core reduces the edible portion of fruit. Core diameters less than 3 cm are usually preferred. Only 12 offspring of H3 x Mauritius backcross and 14 offspring of H4 x Mauritius backcross had cores less than 3.0cm in diameter.

5. Stalk length (cm)

Stalk (peduncle) length of fruits produced by offspring obtained from H3 x Mauritius and H4 x Mauritius backcrosses varied from 12.0 cm to 12.9 cm and 12.0 cm to 12.7 cm respectively and all the fruits of both crosses had strong peduncle with medium height. A strong stalk with medium height is needed to avoid lodging and sun burn.

6. Eye depth, peel thickness and eye shape

Eye depth and peel thickness of fruits produced by offspring derived from H3 x Mauritius backcross varied from 0.7 cm to 1.7 cm and 0.5 cm to 1.5 cm respectively. Similarly eye depth and peel thickness of fruits produced by offspring derived from H4 x Mauritius backcross varied from 0.8 cm to 1.7 cm and 0.8 cm to 1.5 cm respectively. Eye depth and peel thickness of fruits produced by offspring derived from both backcrosses had less eye depth and less peel thickness than that of their parents H3, H4 and Mauritius. Six offspring of H3 x Mauritius backcross and 16 offspring of H4 x Mauritius back cross produced fruits with broad shape eyes. Eye depth, peel thickness and eye shape are also affect the edible portion of the fruit. Broad shallow eye with less peel thickness is more beneficial than characters of projected deep eye with higher peel thickness because considerable amount of flesh removed during the peeling of fruit.

7. Number of fruit-lets

Number of eyes of fruits produced by offspring derived from H3 x Mauritius backcross and H4 x Mauritius backcross varied from 54 to 136 and 72 to 199 respectively. Pineapple fruit composed of 50 - 200 fruit-lets (Eyes) and the fruit-let number depends on the variety (Coppens d'Eeckenbrugge and Leal, 2003).

Fruit quality parameters

1. pH

The pH values of offspring derived from H3 x Mauritius and H4 x Mauritius backcrosses ranged from pH 2.49 - pH 3.91 and pH 2.83 - pH 4.98 respectively. Three offspring of H3 x Mauritius backcross had no significantly different pH value compared to Mauritius and 10 offspring of H4 x Mauritius backcross had significantly higher pH value than Mauritius pineapple. Fruit pH directly influences the sample color, aroma, flavor, oxidation, microbial and chemical stability (Sadler and Murphy, 2010).

2. Titratable Acidity (TA)

Titratable Acidity of offspring of H3 x Mauritius and H4 x Mauritius backcrosses were varied from 0.58 % - 0.74 % and 0.58 % - 0.90 % respectively. Thirteen offspring obtained from H3 x Mauritius backcross and 11 offspring derived from H4 x Mauritius backcross had significantly lower titratable acidity (TA) values compared with Mauritius. TA is a measure of the total organic acids of the sample being analyzed. Similar to the roll of pH, the organic acids present in foods affect the color, flavor, microbial stability and quality maintenance (Sadler and Murphy, 2010). Fruit TA gives a fairly good prediction of tartness although fruit tartness is strongly affected by presence of sugars (Sadler and Murphy, 2010).

3. Total Soluble Solids (TSS) - °Brix

The °Brix values of fruits of offspring derived from H3 x Mauritius and H4 x Mauritius backcrosses ranged from 10.0 - 18.4 and 10.0 - 24.0 respectively. Two offspring of H3 x Mauritius backcross and 8 offspring of H4 x Mauritius backcross had significantly higher °Brix values than their parents. The total soluble solids (TSS) is one of the important qualitative parameter of the fruit (Singleton and Gortner, 1965) and increasing trend of TSS related to the development of taste and flavor in a fruit which make them palatable (Rahman *et al.*, 1979). TSS provides a good indication of sugar content in the sample as these soluble solids are primarily sugars - sucrose, glucose, and fructose.

4. Sweetness Index (SI) - Total Soluble Solids / Titratable Acidity ratio

The flavor and quality of fruits is determined by the interaction between sugar and acid content. Hence, the Sweetness Index (SI) is commonly used for describing the flavor of fruits rather than sugar or acid alone (Potter and Hotchkiss, 1998; Sadler and Murphy, 2010). The Sweetness Index (SI) values of offspring of H3 x Mauritius and H4 x Mauritius backcrosses ranged from 15.38 to 26.66 and 14.40 to 39.34 respectively. In this study, three offspring derived from H3 x Mauritius backcross and 8 offspring derived from H4 x Mauritius backcross produced fruits with significantly higher SI than their parents.

5. Peel and flesh color

Peel color and flesh color of male parent (Mauritius pineapple) fall in the Yellow - Orange group. But both female parents (H3 and H4) fall into Yellow group. Both peel and flesh color of fruits produced by nine offspring of H3 x Mauritius backcross and twelve offspring of H4 x Mauritius backcross grouped into Yellow - Orange group. Peel and flesh color of pineapple fruits is one of the most important qualitative attributes. It is one of the most important criteria for the consumer to purchase fruits from the market. Pineapple with golden color peel and flesh (Yellow - Orange Group) has high demand in both local and export market.

6. Juice content

The percentage of juice in the fruit flesh varied among different varieties. Mauritius variety having medium amount of juice content (44.6 %) and female parents of both crosses have high amount of juice content (H3 - 70.3% and H4 - 80.3%). Varieties with a high juice content (> 60 %) are only suitable for processing and fruits with medium amount of juice (40 % - 60 %) are suitable for fresh fruit and also for processing. The percentage of juice content of offspring of H3 x Mauritius and H4 x Mauritius backcrosses varied from 35 % - 82 % and 22.3 % - 81 % respectively. Seven offspring from H3 x Mauritius backcross and 9 offspring from H4 x Mauritius backcross considered as having medium amount of juice content.

7. Spiny Distribution

In the backcrosses, Mauritius was the male parent while H3 and H4 hybrids were the female parents. Both H3 and H4 hybrid parents only had spines over less than 2 cm from the tip. Mauritius parent had completely spiny leave. Various distribution patterns of spines on leaf margins were observed in offspring obtained from both backcrosses - H3 x Mauritius and H4 x Mauritius. Their spines were located at the tip, base, or along the margins of leaves. According to the spiny distribution pattern, offspring were separated into 4 distinct classes such as Spineless but spines were observed only at the tip (2 - 6 cm from the tip), Spineless but spines observed both at the tip and the base, Uneven distribution of spines along the margin, Spines distributed along the whole leaf margin (Completely spiny). Less number of spines is a favorable character for field practices and fruit handling. Seven offspring derived from H3 x Mauritius backcross and nine offspring from H4 x Mauritius backcross had spineless leaves except at the tip and spines located 2 – 6 cm length along the margins at the tip.

CONCLUSION

Three offspring derived from H4 x Mauritius backcross were selected with desirable fruit quality characters along with other important plant and fruit characters. These elite offspring can be used as breeding material for future pineapple fruit quality improvement programs.

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Services

The listings below are provided as a convenience to readers and should in no way be construed as an endorsement of those providing commercial or professional services.

Commercial Services

Maintain CF 125 continues to be available for use in pineapple plant propagation anywhere in the world. Supplies can be obtained from Repar Corporation, 8070 Georgia Ave., Suite 209, Silver Spring, MD 20910. Tel: (301) 562 – 7330; Fax: (202) 223 – 0141; On the web at www.reparcorp.com; E-Mail: mandava@compuserve.com.

Professional Services

Not updated. Please see Pineapple News No. 25 for listing.

New References on Pineapple

The list below includes papers related to various aspects of pineapple culture, physiology, processing, preservation or byproducts that were published or located for the period since the last issue up to about March 31, 2018. Some papers may seem relatively unrelated to pineapple but the list follows the principle of inclusion to provide the widest possible content. Often, abstracts of the papers listed below can be found on-line. I suggest searching using the paper title. Of course all abstracts of papers published in Acta Horticulturae are available from info@ishs.org. For a larger view, adjust the magnification in Adobe Reader.

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